Effect of meloxicam administration on piglet exploratory behavior and stress post-castration

Honors Research Thesis

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ABSTRACT:

Implementing non-steroidal anti-inflammatory drugs (NSAIDs) such as meloxicam at the time of castration can mitigate pain experienced at the time of castration and following the procedure. Previous research demonstrates that castration is painful and more research is needed to make a change in the industry to incorporate pain relief protocols during castration. The objective of this study was to assess the impact of meloxicam administration on piglet exploratory behavior and stress post-castration.

Piglets (n=19) were enrolled on the trial at 13 to 15 days of age and underwent a two-week acclimation period. Piglets were randomly assigned to one of two treatments: 1) control and 2) Meloxicam (1.0 mg/kg PO). At baseline, piglets were transported individually to the testing room and behavior was evaluated for three minutes using live observation. On test day, piglets received treatment and castrated approximately 15 minutes post treatment administration. Castrated piglets were then transported to the testing room individually and piglet behavior was evaluated for three minutes. There was no difference between the latency to touch an object or researcher pre and post-castration ($P > 0.05$). Regardless of treatment, there was a 41.8 ± 33.6 second (Mean ± SD) latency for the piglet to touch an object and a 129 ± 28.6 second (Mean ± SD) latency for the piglet to touch the researcher. Regardless of treatment, there was a 50.3 ± 50.3 second (Mean ± SD) latency to touch an object and a 107.7 ± 51.8 second (Mean ± SD) to touch the researcher. There was no treatment effect on the latency to touch an object or researcher in the pre or post-castration behavioral assessments ($P>0.05$). Additionally, there was no difference between pre-castration and post-castration thromboxane B2 levels ($P > 0.05$). Treatment had no effect on thromboxane B2 levels (Table 3).

INTRODUCTION:

Castration is a procedure commonly performed in the US swine industry on male piglets. This procedure is painful as demonstrated by changes to the behavior and physiology of the piglet including decreased nursing time, weight loss, increased tendency to fight with litter mates, and increased vocalization. Although to date, castration is performed without an anesthesia or analgesia, research has demonstrated that pain relieving drugs such as Non-steroidal anti-inflammatory drugs (NSAID) can effectively mitigate pain associated with castration. To quantify pain severity in farm animals, behavioral trials can be performed to assess the deviations in natural behavior and physiological response between piglets that received a form of pain relief and those who did not. The objective of this research project was to determine the effect of meloxicam administration on exploratory behaviors in weaned piglets undergoing castration.

MATERIALS AND METHODS:

The Ohio State University Institutional Animal Care and Use Committee approved the protocol used throughout this study. Furthermore, all piglets included in this experiment were cared for in accordance with the Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching.

Nineteen non-castrated male piglets from the Ohio State Don Scott Swine Facility in Columbus, Ohio, USA were used in this study during March 2018. Prior to the beginning of this study, all piglets were housed in a farrowing crate (5 feet by 7 feet) and remained with their littermates and sow. Acclimation of the piglets began at approximately three weeks before weaning and the trial ended a week after the piglets were weaned. The litters of piglets were selected to be 13 to 15 days old at the start of the trial, have no apparent health problems, and

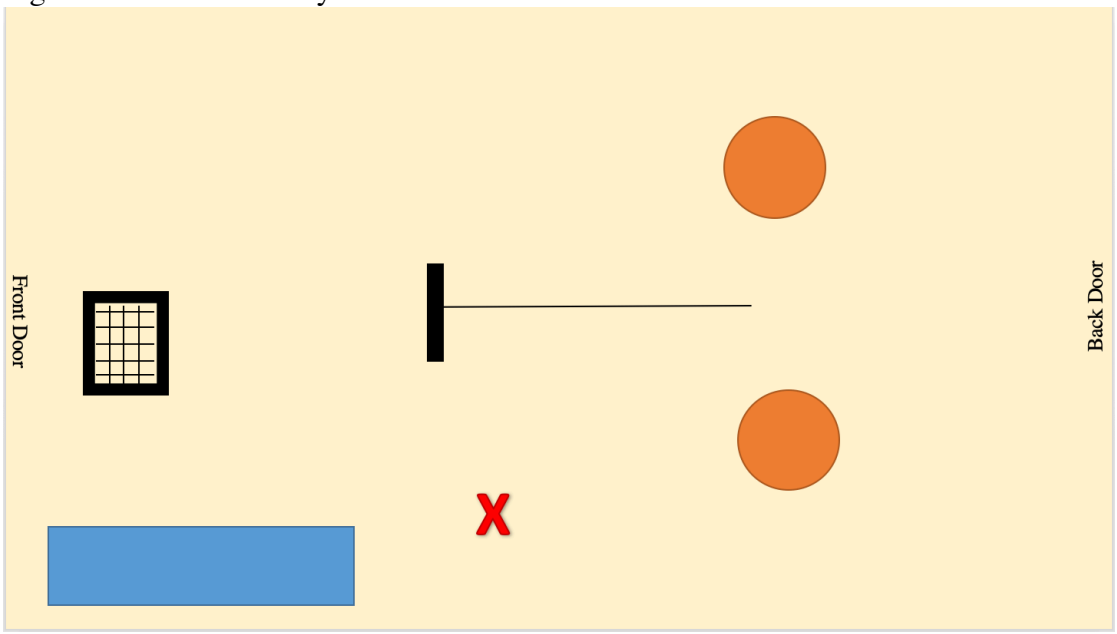
have four to ten males. Piglets were weaned at 27 to 29 days of age and transported to nursery facilities where they were housed in groups of four with littermates. Pens (4 feet by 4 feet) had a grated flooring, a conventional dry feeder with an adjustment plate, and ad lib access to a water pipe that had two nozzles.

Acclimation period

Prior to weaning, piglets were acclimated to the researcher through direct contact with researcher as the researcher sat inside the farrowing crate and encouraged interactions with the piglets. The researcher remained in each farrowing crate for fifteen minutes for 14 days until the day of weaning. Positive reinforcement via food reward was used and the piglets exhibited play behavior and excitement in the farrowing crates.

For two days following weaning weaned pigs were acclimated to the testing room in groups of four. Each group had five minutes to explore the room on each day (Fig 1).

Figure 1. Trial Room Layout



The room consisted of the researcher (red X), storage objects (blue rectangle), two orange buckets (orange circles), a broom (black broom), and a grate by the front door (black grate).

Following group acclimation, three days were allotted for room acclimation on the individual piglet level. Each piglet was placed in the room by itself and was able to remain in the room for three minutes each day.

Treatment and castration procedure

Piglets were randomly assigned to one of two treatments:

1) Control (n= 10 piglets)

2) Meloxicam (1.0 mg/kg PO; n=9 piglets).

Treatments were administered 15 minutes prior to castration was performed. Castration was performed by two licensed veterinarians and was done using a scalpel blade. All piglets received a local anesthetic prior to castration, followed by meloxicam treated pigs receiving an oral dose of meloxicam and control treated pigs receiving no NSAID treatment.

Behavioral assessment

After the acclimation period was complete, baseline behavioral assessments (Pre-castration) were conducted the day prior to castration for all piglets. Behaviors evaluated are described in Table 1. The same behavioral assessment was conducted on the day of castration after treatments were administered (post-castration). Each individual piglet had three minutes to explore the room and play with the objects or researcher (baseline and test day). The time the piglet took to contact the objects or researcher was recorded in addition to the amount of times the piglet returned to interact with the objects or researcher. The researcher in the room was blind to the treatments of the piglets and was not present for the castration or medication of the piglets (Table 2).

Table 1. Behavioral ethogram

Behavior	Description
Play	The piglet actively interacts with the objects or researcher for more than three seconds. The piglet does not exhibit any fear behavior or fear vocalizations but may have excitatory vocalizations. The piglet's focus was on playing, not leaving the room.
Fear	Piglets that stood in one spot without moving, piglets actively looking to escape the room, and piglets that consistent exhibited fearful vocalizations were recorded as exhibiting fear behavior.
Vocalizations	The amount and type of vocalizations were noted to show play or fear behavior. High pitched, consistent vocalizations denoted fearful vocalizations. Squeals intermittent to interacting with an object or researcher denoted excitatory vocalizations.
Exploratory	Piglets focused on exploring the room, nibbling on different objects or the researcher, and piglets that interacted with the objects spent majority of their time exploring the room.

Table 2. Experimental timeline

Trial days 1-14	Acclimation to researcher in farrowing crate for fifteen minutes per litter
Trial day 15	Piglets weaned (Age: 27-29 days)
Trial days 16-17	Group acclimation to trial room for five minutes per group
Trial days 18-20	Individual acclimation to trial room for three minutes per pig
Trial day 21	Pre-castration behavioral assessment for three minutes per pig
Trial day 22	Post-castration behavioral assessment for three minutes per pig

Thromboxane B2 Analysis

Samples were analyzed using Cayman Chemical 11-dehydro thromboxane B2 Competitive ELISA (Cat #519510). Plasma samples were extracted after thawing at 22^{o}C and aliquoting 500 uL of the sample into a clean glass tube. Two mL of cold acetone was added to each tube and centrifuged at 2000 x g for 20 minutes. The acetone layer was removed and placed

into a labeled borosilicate glass tube and dried under a nitrogen stream at 40^{o}C. The sample was reconstituted in 250 uL of assay buffer, then vortexed and centrifuged at 215 x g for 7 minutes. Samples were added to wells as indicated by the ELISA manual. Sample were measured at a 405 nm absorbance and analyzed using a four-parameter logistic regression fit.

RESULTS:

There was no difference between the pre-castration and post-castration on the latency to touch an object or the researcher (P> 0.05). Pre-castration behavioral assessment demonstrated a 41.8 ± 33.6 second (Mean ± SD) latency for the piglet to touch an object regardless of treatment and a 129 ± 28.6 second (Mean ± SD) latency for the piglet to touch the researcher. Post-castration behavioral assessment demonstrated a 50.3 ± 50.3 second (Mean ± SD) latency to touch an object regardless of treatments and a 107.7 ± 51.8 second (Mean ± SD) latency to touch the researcher. There was no treatment effect on latency to touch an object or researcher in either the pre or post-castration behavioral assessment (P> 0.05).

There was no difference between pre-castration and post-castration thromboxane B2 levels (P> 0.05). In addition, treatment had no effect on thromboxane B2 levels (Table 3).

Table 3. Thromboxane B2 Levels (pg/mL) Pre and Post-castration for Meloxicam and Control Treated Piglets

Treatment	Pre-castration	Post-castration
Control (n=10)	186.0 ± 67.8	216.0 ± 113.8
Meloxicam (n=9)	162.7 ± 55.3	163.0 ± 51.3

DISCUSSION:

Throughout the United States, major swine producers castrate piglets to prevent boar taint from creating an unpleasant taste and smell, depreciating the value of the pork (Swine Care Handbook, 2018). Piglets are castrated without any form of pain relief as many argue the pain

mitigating medications are not practical in the industry (Swine Care Handbook, 2018). Doing so violates the welfare of piglets as they are not free from pain or discomfort; one of the five freedoms utilized to assess individual animal welfare (Swine Care Handbook, 2018). Discerning pain is not an easy task as animals do not have the ability to communicate with humans. Instead, using blood tests to analyze cortisol levels, a stress hormone released from the adrenal cortex, and evaluating deviations in normal pig behavior can be used as indirect indicators to quantify pain. The objective of this research project was to determine the effect of meloxicam administration on exploratory behavior and stress in weaned piglets undergoing castration.

Previous research has demonstrated that castration pain is associated with deviations to a piglet's physiological response and behavior. Work conducted by Taylor and colleagues (2013) demonstrated castration pain results in changes to piglet's vocalization and decreases in several behaviors including less time lying down, more time standing or sitting, and older piglets spent less time nursing. Kluivers-Poodt and colleagues (2013) also demonstrated an increase in pain specific behaviors (i.e. tail wagging, stiffness, and huddled up posture) immediately post-castration. These behaviors can be decreased when piglets receive analgesics such as meloxicam prior to castration (Kluivers-Poodt *et al*., 2013) In contrast to these studies and other, our study did not demonstrate a difference in behaviors between piglets treated with meloxicam compared to non-treated piglets. The latency to touch an object or researcher was not different between groups post-castration, nor was it different within group when comparing the pre-castration and post-castration behavior period.

Future work in this area would benefit from increasing the total number of piglets enrolled and including more replicates of the study. In addition, acclimating piglets to the veterinarian and those assisting with castration may be beneficial to minimize stress associated

with a novel human which may have influenced their behaviors. Finally, longer observation periods may have allowed for a better evaluation of pig exploratory behavior. Future studies should consider increasing the time the pig has to explore the environment as well as consider evaluating pigs in pairs to decrease stress associated with isolation from littermates.

Although there was no difference between pre-castration and post-castration thromboxane B2 and treatment had no effect on thromboxane B2 levels, the blood work does show the importance of pain mitigation during castration. When analyzing the numbers for the groups, the mean thromboxane B2 levels in the meloxicam treated piglets remained constant pre and post-castration. The mean thromboxane B2 levels in the control piglets did not remain constant, and increased by 30 pg/mL. Considering that the number of piglets enrolled in this study was low and only ten piglets were assigned to each treatment group, it is not surprising that there was not a difference in the levels of thromboxane B2. Repeating the study with a larger number of piglets assigned to each treatment group may yield significant results.

CONCLUSION:

The objective of this research project was to determine the effect of meloxicam administration on exploratory behavior and stress in weaned piglets undergoing castration. Pigs that received meloxicam prior to castration did not show a difference in exploratory behavior as defined by latency to touch novel objects or a human as compared to non-treated piglets. Future research is needed to refine behavioral sampling methodologies and to evaluate the efficacy of other analgesics drugs on exploratory behavior of castrated pigs.

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